BEST AVAILABLE IMAGES

Defective images within this document are accurate representations of the original documents submitted by the applicant.

Defects in the images may include (but are not limited to):

- BLACK BORDERS
- TEXT CUT OFF AT TOP, BOTTOM OR SIDES
- FADED TEXT
- ILLEGIBLE TEXT
- SKEWED/SLANTED IMAGES
- COLORED PHOTOS
- BLACK OR VERY BLACK AND WHITE DARK PHOTOS
- GRAY SCALE DOCUMENTS

IMAGES ARE BEST AVAILABLE COPY.

As rescanning documents *will not* correct images, please do not report the images to the Image Problem Mailbox.

REMARKS

Claims 59-64 are pending in the subject application.

Rejection under 35 U.S.C. § 102(b) - Eisenberg

On page 2 of the January 14, 2004 Office Action the Examiner maintained the rejection of claims 59-61 and 63 under 35 U.S.C. §102(b), as allegedly anticipated by Eisenberg. The Examiner stated that Eisenberg teaches oxandrolone orally administered at 1.25 to 20 mg, encompassing the dosages claimed in the application.

On page 4 of the Office Action, in response to applicant's prior remarks about this rejection, the Examiner alleged that Eisenberg clearly stated those oxandrolone dosages meeting the dosages envisioned by the application, thereby providing a clear anticipation for the presented claims. The Examiner further alleged that with regard to the administration levels taught by the prior art, the skilled artisan would assume if Eisenberg administered the medicament in multiple doses, the instant regimen would have been disclosed. The Examiner also stated that he assumes the underlying article by Eisenberg would clarify the issue raised by the Applicant, and that if the instant application is appealed, the Eisenberg article will be obtained.

In response, applicant maintains that the Eisenberg article (hereinafter "Eisenberg") does not anticipate the subject claims because Eisenberg does not disclose or anticipate a pharmaceutical composition comprising oxandrolone and a pharmaceutical carrier, wherein the oxandrolone is present in an amount of 7.5 mg or more. For the convenience of the Examiner, applicant provides a copy of Eisenberg as **Exhibit A**

and respectfully points out that the Chemical Abstracts summary of Eisenberg (hereinafter "the abstract") has apparently lead the Examiner to incorrectly understand that the hormones listed on lines 4 and 5 of the abstract were all administered at each respective dosage listed in parentheses in line 5. In contrast, inspection of Eisenberg at page 567 (first column, lines 12-22) indicates that in fact the hormones were each administered at a single dosage, and oxandrolone in particular was administered only at 5 mg per day (line 20). Eisenberg does not mention any dosage of oxandrolone other than 5 mg per day, and moreover does not provide information regarding administrative forms or pharmaceutical compositions comprising oxandrolone. Furthermore, Eisenberg does not teach or disclose a pharmaceutical composition comprising oxandrolone and certainly not a pharmaceutical composition in which the oxandrolone is present in an amount of 7.5 mg or more. Thus, the subject claims are novel over Eisenberg.

Rejection under 35 U.S.C. § 103 - Pike et al. And Eisenberg

On page 3 of the January 14, 2004 Office Action the Examiner maintained the rejection of claims 59-64 under 35 U.S.C. §103, as allegedly unpatentable over Pike et al. and Eisenberg. The Examiner alleged that Pike et al. and Eisenberg teach the claimed oxandrolone compositions as old and well known in combination with various pharmaceutical carriers and excipients in a dosage form, and that the medicament is taught as useful for treating various maladies. The Examiner noted that claims 59-64 and the primary references differ as to 1) the employment of these medicaments by conventional means and routes, and 2) administration of the medicaments for specific therapeutic goals. The Examiner further noted that claims 59-

64 specifically require conventional pharmaceutical composition and delivery routes, and alleged that Pike et al. (Claims 1-8, and column 4, lines 35-36) and Eisenberg employed the claimed oxandrolone in a dermal form, subcutaneously, IM, transdermally and orally, not specifying the exact formulation. The Examiner alleged that the skilled artisan would have seen conventional compositions, and the administration of these compositions by conventional means as residing in the skilled artisan purview. The Examiner referred to In re Dillon (CAFC 1990) and stated that the court sitting en banc ruled that the recitation of a new utility for an old and well known composition does not render that composition new.

In response, applicant reiterates the argument presented in the October 9, 2003 communication that Pike et al. is not prior because the filing date of Pike et al. is subsequent to the effective filing date of applicant's claimed invention.

Specifically, applicant's claims are entitled to the October 20, 1992 filing date of U.S. Serial No. 07/963,469 which has a specification identical to that of the instant application. A copy of U.S. Serial No. 07/963,469 is attached as **Exhibit B** for the Examiner's convenience. The Declaration and Power of Attorney submitted as Exhibit A to applicant's October 9, 2003 communication contains a claim to benefit of the October 20, 1992 filing date of U.S. Serial No. 07/963,469. Finally, the subject specification contains a cross-reference to U.S. Serial No. 07/963,469. Accordingly, the subject application satisfies the requirements to be accorded benefit of the October 20, 1992 filing date of U.S. Serial No. 07/963,469.

Applicant notes that the filing receipt issued by the U.S. Patent Office in connection with of the subject application omits a reference to U.S. Serial No. 07/963,469 filed October 20, 1992. This is an error. Therefore, applicant has filed concurrently with this Amendment a Request To Correct Error In Filing Receipt, a copy of which is attached as **Exhibit C**.

In view of the above, and on the basis of the documents previously submitted, applicant respectfully requests accordance of October 20, 1992 as the effective filing date of the subject application.

Upon entry of October 20, 1992 as the effective filing date of the instant application, Pike et al. falls away as a prior art citation because the relevant disclosure of Pike et al. is not found in Pike's parent or grandparent applications in its priority chain. Specifically, Pike et al. is U.S. Patent No. 5,640,586 which issued from U.S. Serial No. 62,886, filed May 17, 1993, which in turn was a continuation-in-part of U.S. Serial No. 952,513 filed February 3, 1993, which in turn was a continuation-in-part of U.S. Serial No. 684,612 filed April 12, 1991 and which is now U.S. Patent No. 5,211,952. The February 3, 1993 filing date of U.S. Serial No. 952,513 is subsequent to the October 20, 1992 effective filing date of the instant application. The immediately preceding April 12, 1991 filing date of U.S. Patent No. 5,211,952 is prior to that of the instant application, but Pike's U.S. Patent No. 5,211,952, has a specification different from that of Pike et al. currently being cited. Specifically, Pike's U.S. Patent No. 5,211,952 does not disclose oxandrolone at all. A copy of Pike's U.S. Patent No. 5,211,952 is attached as **Exhibit D** for the Examiner's convenience. In this regard, applicant refers

to MPEP 2136.03 IV which states:

> In order to carry back the 35 U.S.C. 102(e) critical date of the U.S. patent reference to the filing date of a parent application, the parent application must (a) have a right of priority to the earlier date under 35 U.S.C. 120 or 365 (c)and (b) support the invention claimed as required by 35 U.S.C. 112, first paragraph. "For if a patent could not theoretically have issued the day the application was filed, it is not entitled to be used against another as "secret prior art" under 35 U.S.C. 102(e). In re Wertheim, 646 F.2d 527, 537, 209 USPQ 554, 564 (CCPA 1981)..."

Without Pike et al. as a reference, the rejection under 35 U.S.C. § 103 is moot.

Supplemental Information Disclosure Statement

In accordance with their duty of disclosure under 37 C.F.R. §1.56, applicant points out that U.S. Serial No. 09/469,817, filed December 22, 1999 and previously disclosed to, and considered by, the Examiner has issued as U.S. Patent No. 6,670,351. Applicant attaches a Form PTO-1449 as **Exhibit E** listing U.S. Patent No. 6,670,351 for the Examiner to initial and return to applicant.

A copy of U.S. Patent No. 6,670,351 is attached hereto as **Exhibit F.**

If a telephone interview would be of assistance in advancing prosecution of the subject application, applicant's

undersigned attorney invites the Examiner to telephone him at the number provided below.

No fee is deemed necessary in connection with the filing of this Amendment. However, if any other fee is required, authorization is hereby given to charge the amount of any such fee to Deposit Account No. 03-3125.

Respectfully submitted,

Gary J. Gershik

John P. White
Registration No. 28,678
Gary J. Gershik
Registration No. 39,992
Attorneys for Applicant
Cooper & Dunham LLP
1185 Avenue of the Americas
New York, New York 10036
(212) 278-0400

I hereby certify that this correspondence is being deposited this date with the U.S. Postal Service with sufficient postage as first class mail in an envelope addressed to: Mail Stop AF, Commissioner for Patents, P.O. Box 1450, Alexandria, VA 22313-1450.

Gary J. Gershik 3/5/04

John P. White Date
Reg. No. 28,678
Gary J. Gershik
Reg. No. 39,992

Effects of Androgens, Estrogens and Corticoids on Strontium Kinetics in Man[1]

EUGENE EISENBERG

The Department of Medicine, University of California, Medical Center, San Francisco, California

ABSTRACT. The effects of estrogens, androgens and corticoids on kinetics of stable strontium metabolism were evaluated. The rapidly miscible pool volume, bone deposition rate and urinary excretion rate were determined before and after treatment of 87 subjects with androgens and estrogens, 44 subjects with corticoids and 34 subjects with placebos. Androgens and estrogens decreased urinary excretion rates and estrogens tended to decrease bone deposition. They thus appear to be anticatabolic for bone instead of anabolic, as previously thought. Estrogens may also be anti-anabolic. Corticoids did not decrease bone deposition rates, but did uniformly increase urinary excretion rates. They thus do not appear to be anti-anabolic for bone, as previously thought. Correction of bone deposition rates for fecal excretion rates did not affect the results. These studies did not reveal whether the changes in urinary excretion rates induced by both gonadal and glucocorticoid steroids were attributable to effects on the kidney, on the bone, or both. (*J Clin Endocr* 26: 589, 1966)

BALANCE studies on men and women treated with androgens and estrogens show that these hormones increase retention of calcium, phosphorus and nitrogen (1, 2). This fact has been used as evidence to support the thesis that these agents stimulate new bone formation in man. More direct measurements suggest that they do increase new bone formation in other species (3–7), but, even after years of therapy with androgens and estrogens, convincing evidence of new bone formation in adult patients has not been shown by anatomic or radiographic means. Treatment with glucocorticoids, in contrast, clearly causes both negative calcium balance and loss of bone mass in human adults (1), as well as in mature animals of other species. The physical appearance of bone from animals and patients exposed to the effects of glucocorticoids for long periods has been interpreted to mean that glucocorticoids produce loss of bone tissue by decreasing deposition of new bone (6–9). Hence, steroids are believed to induce both positive and negative changes in calcium balance by direct effects on the rate of bone deposition.

Analysis of the behavior of stable strontium has revealed that differences in its rate of skeletal deposition in various metabolic diseases (10) can be detected. Therefore, kinetic studies have been done in subjects before and during treatment with steroid hormones in an effort to demonstrate the supposed effects of these agents on bone deposition rate.

Materials and Methods

The effects of estrogens and androgens were studied in 57 women with postmenopausal osteoporosis, 8 men with senile osteoporosis, 11 men with idiopathic osteoporosis and 3 elderly men without osteoporosis. The effects of corticoids were measured in 21 men and women having diseases requiring large doses of corticoids and in 23 normal male volunteers. Intestinal excretion rate of strontium was measured in 29 of the 44 subjects who received corticoids.

Received December 22, 1965; accepted February 15, 1966.

This work was supported in part by funds allocated by the Committee on Research and the Academic Senate Committee on Research, University of California, School of Medicine, San Francisco, Lederle Laboratories, Merck Sharp & Dohme, G. D. Searle & Co., E. R. Squibb & Sons and The Upjohn Company.

[1] Preliminary reports of this work have been published (11, 12).

The inherent variability of the method was determined by replicate kinetic studies in 34 subjects who had placebo therapy between the first and second tests. Each subject was asked to avoid major fluctuations in calcium intake by eliminating milk and cheese from his diet for the duration of the study. Strontium kinetics were determined on each subject before any therapy. The experimental subjects were then treated with one of the hormones, and kinetic studies were repeated during the fifth or sixth week of therapy. The hormones and the daily oral doses used were: cortisol 80–120 mg, prednisone 20–30 mg, triamcinolone 12–18 mg, 6α-fluoroprednisolone 12 mg, dexamethasone 3.0 mg, 6α-fluorotriamcinolone 24 mg, conjugated equine estrogens 2.5 mg, ethinyl estradiol 0.1 mg, methallenestrol 9 mg, 16α-methyl estriol-16β, 17β-3-methyl ether 20 mg, fluoxymesterone 10 mg, oxandrolone 5 mg, norethandrolone 20 mg, oxymetholone 7.5 mg, 7,17-dimethyl testosterone 1.25 mg. Three androgens were given by intramuscular injection: testosterone enanthate 200 mg every 2 weeks, testosterone caprinoyl acetate 200 mg every 2 weeks, nandrolone phenopropionate 50 mg every 2 weeks.

The kinetic analysis and the chemical methods for serum and urine strontium determinations have been published previously (10). The kinetic method is based on the single compartment, "turnover difference" concept. Ten mEq of nonradioactive strontium is given intravenously. Twenty-four hr urine collections and daily blood samples are taken for 6 days. Miscible strontium pool size and its turnover rate are calculated by standard dilution formulas. Urinary excretion rate is calculated from an integration of the serum clearance curve and urinary strontium content. The rate of bone deposition of strontium is taken as the difference between the total turnover rate and urinary excretion rate. Numerous studies suggest that rate of bone deposition of calcium is very close to that of strontium (13–15), and recently Auberi and associates (16) concluded that strontium is a valid qualitative marker for calcium. Strontium is lost from the miscible pool via the sweat glands, but in most cases this loss is negligible. Since the subject's activity remained constant and there is little or no change in ambient temperature and humidity in San Francisco, it was believed that sweat loss would be identical during the 2 kinetic studies in each subject and it was therefore not measured routinely. Stool specimens were collected from the day of strontium infusion until 24 hr after urine collections were completed. The stools were homogenized in 9 times their weight of distilled water. A 5 ml portion of the homogenate was digested with 2 ml of 60% perchloric acid and 1 drop of selanium oxychloride. The clear digest was diluted to 25 ml with a solution containing 1.33N HCl, 0.755 mM KH_2PO_4 and 27.45 mM Na_2CO_3. The strontium concentration was then determined in the flame spectrophotometer by reading between appropriate standards. Strontium, 0.02 mEq, was added to 5 ml samples of each stool homogenate in order to determine the degree of recovery of strontium. All determinations were done in duplicate, and the results of duplicate analyses always agreed within 3%.

Accurate separation of feces into those formed during and after the mixing phase is not possible, as it is with urine. To permit calculation of the intestinal clearance rate of strontium it was assumed that the ratio

$$\frac{\text{strontium excreted following mixing phase}}{\text{total strontium excreted from time of injection}}$$

was the same for feces as it was for urine. The following formulas were used:

$$1.\ \frac{F_{1-t}}{F_{0-t}} = \frac{U_{1-t}}{U_{0-t}}$$

$$2.\ F^* = \int_1^t dF\delta_0 \int_1^t d^{-kt}dt = [F_{(1-t)}]k/B_1 - S_1$$

F_{1-t} = fecal strontium excretion from end of first 24 hr to completion of collection; F_{0-t} = total fecal strontium content; U_{1-t} = urinary strontium from end of first day to end of fifth; U_{0-t} = total urinary strontium content; F^* = fecal excretion rate; k = total miscible pool turnover rate constant; S_1 = theoretical serum level at time of injection; S_t = serum level at end of first 24 hr; S_t = serum level at end of collection.

Results

The calculated rapidly miscible pool, its turnover rate and the rates of urinary excretion and bone deposition (Table 1) were similar to those previously obtained in normal subjects, athletes and osteoporotics in this laboratory (10). When these groups had a second test done after six weeks of placebo therapy, some spontaneous variability in the kinetic measurements was noted, but the changes were not significant (Table 1). The maximum mean variability of any measurement was 6%.

Since each treatment group included subjects with various diagnoses as well as some without demonstrable disease, the data in most groups had a wide range. The scatter tended to mask the effects of treatment

TABLE 1. Reproducibility of strontium kinetics

Group	No.	Urinary calcium mEq/24 hr		Miscible pool liters		Total turnover liters/24 hr		Urinary excretion liters/24 hr		Bone deposition liters/24 hr	
		C	T	C	T	C	T	C	T	C	T
Normal subjects	10	6.63 ±0.60	6.30 ±0.70	46.25 ±0.88	46.64 ±2.3	14.24 ±0.81	14.46 ±0.93	3.65 ±0.14	4.44 ±0.16	10.48 ±0.54	9.97 ±0.83
Athletes	10	10.60 ±0.63	10.67 ±1.10	50.71 ±2.45	50.62 ±2.30	20.21 ±1.08	21.54 ±1.09	7.32 ±0.71	8.72 ±0.92	18.91 ±0.96	16.20 ±1.17
Pneumoencephalogram Osteoporosis	14	4.68 ±0.30	4.15 ±0.59	29.63 ±1.44	32.16 ±1.10	8.05 ±0.45	8.11 ±0.60	2.29 ±0.23	2.10 ±0.25	6.34 ±0.34	6.01 ±0.83

Figures are mean ± standard error.
C represents pretreatment measurement; T represents measurement after 6 weeks of placebo therapy.

when the actual values before and after treatment were compared. To clarify the effects of therapy in a physiologically nonuniform group, each subject was used as his own control. Therefore, the data presented in Tables 2, 3 and 4 are the changes induced by each agent. No differences in response of strontium kinetics to therapy could be discerned between normal subjects and patients; hence, the effects of each therapy are shown for the entire group without regard to diagnosis.

Androgens (*Table 2*). None of the agents used produced a significant change in the size of the miscible pool. The rate of total turnover of the miscible pool was significantly diminished. This was attributable to a decrease in the rate of urinary excretion of strontium. There was no significant alteration in bone deposition rate. The decreases in urinary excretion rate of strontium from its miscible pool were paralleled by decreases in urinary calcium excretion.

Estrogens (*Table 3*). Estrogen therapy reduced the urinary excretion rate of strontium (and urinary calcium excretion) without affecting the miscible pool size. In contrast to the results with androgen, there

TABLE 2. Change in urinary calcium excretion and in strontium kinetics induced by six weeks of androgen therapy

	No.	Urinary calcium mEq/24 hr	Miscible pool liters	Total turnover rate liters/24 hr	Urinary excretion rate liters/24 hr	Bone deposition rate liters/24 hr
Testosterone Enanthate	7	−0.80 ± 0.80	+0.60 ± 1.71	−1.06 ± 0.81	−0.87 ± 0.80	−0.19 ± 0.88
Fluoxymesterone	10	−0.80 ± 0.20	+1.84 ± 1.00	−0.84 ± 0.42	−0.38 ± 0.09	−0.18 ± 0.11
Cantrolone	10	−1.30 ± 0.71	−0.80 ± 1.31	−1.39 ± 0.37	−1.15 ± 0.40	−0.72 ± 0.60
Oxymetholone	18	−2.00 ± 0.60	+1.30 ± 1.84	−2.02 ± 0.74	−2.81 ± 0.48	+0.22 ± 0.66
Nandrolone Phenopropionate	5	−3.12 ± 0.75	+0.78 ± 4.18	−2.08 ± 1.66	−1.48 ± 1.11	−1.40 ± 0.04
7,17-Dimethyl Testosterone	0	−2.20 ± 1.03	−2.85 ± 0.08	−1.03 ± 0.61	−1.10 ± 0.43	+0.40 ± 0.46
Testosterone Caprinoyl Acetate	3	−0.82, −0.80, −0.00	+4.41, +11.00, +3.02	−4.41, −2.03, −3.80	−1.34, −2.46, −2.62	−4.15, −1.34, +0.11
Norethandrolone	2	+0.80, −0.00	−10.81, +2.11	−1.11, −2.62	+0.11, −1.16	−1.10, −3.60

Figures are means of the differences between control and treatment data ± standard error.
For groups with N less than 5, individual values are given.

TABLE 3. Change in urinary calcium excretion and in strontium kinetics induced by six weeks of estrogen therapy

	No.	Urinary calcium mEq/24 hr	Miscible pool liters	Total turnover rate liters/24 hr	Urinary excretion rate liters/24 hr	Bone deposition rate liters/24 hr
Conjugated Equine Estrogen	7	−3.16 ± 1.11	−0.30 ± 1.54	−3.11 ± 0.68	−2.64 ± 0.79	−0.78 ± 0.30
Methallenestril	9	−1.32 ± 0.40	−0.08 ± 1.80	−1.80 ± 0.09	−0.98 ± 0.01	−0.64 ± 0.03
Ethinyl Estradiol	2	−1.62, −1.33	+1.77, −7.85	−8.89, −4.88	−7.04, −1.33	−6.18, −3.52
Methyl Estriol	3	−1.89, −1.08, −0.87	+4.31, +5.22, −0.02	−1.01, −2.81, −0.87	−0.91, −0.84, −0.11	−1.34, −2.32, −0.73

Figures are mean of the differences between control and treatment data ± standard error.
For groups with N less than 5, individual values are given.

was a significant depression of bone deposition rate of about 0.6 liter of miscible pool per 24 hours. Since these were all patients with postmenopausal osteoporosis, this represented roughly 10% decrease in the bone deposition rate.

Glucocorticoids (*Table 4*). In general, significant changes in the volume of the miscible pool were not seen in patients treated with this group of compounds. Most of the corticoids produced an increased rate of turnover, largely due to an increase in the urinary excretion rate. For all corticoid-treated subjects this increase was about 30% above pretreatment excretion rates. As with the androgens and estrogens, roughly parallel changes in urinary calcium excretion were seen. Treatment with cortisol and dexamethasone resulted in increased bone deposition rates; prednisone and triamcinolone did not change the bone deposition rate; 6α-fluorotriamcinolone decreased the bone deposition rate.

The data on fecal strontium excretion are shown in Table 5. Chemical recovery of stool strontium averaged 80.5% and was unaffected by corticoid therapy. The incomplete recovery of strontium from stool homogenates results from the presence of large amounts of phosphate, sulfate and other salts that inhibit excitation of

TABLE 4. Change in urinary calcium excretion and in strontium kinetics induced by six weeks of corticoid therapy

	No.	Urinary calcium mEq/24 hr	Miscible pool liters	Total turnover liters/24 hr	Urinary excretion liters/24 hr	Bone deposition liters/24 hr
Cortisol	5	+2.28 ± 0.38	+4.32 ± 0.30	+3.08 ± 0.41	+1.68 ± 0.19	+1.74 ± 0.28
Prednisone	14	+4.88 ± 0.50	+0.01 ± 3.15	+1.30 ± 0.98	+3.55 ± 0.68	+0.62 ± 0.82
Triamcinolone	6	+3.70 ± 1.00	+4.06 ± 1.70	+3.81 ± 0.81	+3.31 ± 0.60	+0.88 ± 0.90
6α-Fluorotriamcinolone	0	+0.58 ± 1.15	+0.17 ± 1.07	+2.10 ± 1.04	+3.17 ± 0.09	−1.17 ± 0.70
Dexamethasone	7	+3.80 ± 1.05	+0.21 ± 3.30	+0.83 ± 1.13	+1.79 ± 0.70	+1.74 ± 0.03
6α-Fluoroprednisolone	4	+5.81, +5.22, +11.88, +3.41	+3.01, −5.82, −0.32, −0.83	+2.01, +7.61, +3.19, +1.02	+7.81, +5.11, +3.53, +2.22	+3.11, +2.54, −0.40, −0.09

Figures are mean of the differences between control and treatment data ± standard error.
For groups with N less than 5, individual values are given.

TABLE 6. Effects of glucocorticoids on fecal excretion of strontium

No.	% recovery of Sr added to stool homogenate		% of administered dose appearing in 6-day fecal collection		Rate of fecal clearance of Sr from miscible pool		Rate of bone deposition of Sr from miscible pool without fecal correction		Rate of bone deposition of Sr from miscible pool with fecal correction	
	C	T	C	T	C	T	C	T	C	T
					Prednisone					
11	84.5 ±1.1	82.5 ±0.6	5.87 ±0.78	4.24 ±0.59	0.60 ±0.01	0.51 ±0.01	12.3 ±1.34	12.1 ±1.00	11.9 ±1.34	11.4 ±1.10
					6α-Fluoroprednisolone					
9	83.0 ±1.2	80.7 ±3.8	7.50 ±0.80	7.02 ±0.90	1.22 ±0.24	1.22 ±0.17	10.97 ±0.68	11.40 ±1.06	9.74 ±0.81	10.18 ±1.13
					Dexamethasone					
8	73.6 ±4.1	74.8 ±4.3	3.25 ±1.01	8.17 ±0.03	0.09 ±0.21	1.66 ±0.33	11.4 ±0.57	13.2 ±1.24	10.5 ±0.44	11.6 ±1.18
					6α-Fluoroprednisolone					
2	22, 82, 78	73, 70, 84	0.7, 4.1, 3.7	12.7, 8.0, 6.2	0.84, 0.04, 0.87	2.00, 0.80, 1.03	14.1, 0.2, 12.6	10.2, 6.7, 13.3	12.1, 5.0, 12.0	10.1, 7.0, 12.2

Figures are mean ± standard error, except individual data are given for 6α-fluoroprednisolone.

strontium (and calcium) in the flame. The percentage of the dose of strontium excreted in six days via the feces before corticoid therapy ranged from 2.3 to 17.0, with a mean of 6.3. Corticoid therapy caused this value to increase in 15 subjects, and to decrease in nine; in five subjects no change occurred. As expected, the rate of fecal excretion from the miscible pool did not always respond to corticoid therapy to the same degree or in the same direction as the percentage of the dose excreted in feces.

Occasional subjects showed a marked increase in the rate of fecal excretion of strontium when a corticoid was administered; in general, however, there was no significant change in fecal excretion rate of strontium in any corticoid-treated group. When the fecal excretion rate is included in the bone deposition rate, the latter is overestimated by an average of 7% both before and during corticoid therapy. Therefore, measurement of fecal excretion rate did not affect the differences in the rate of bone deposition produced by corticoid therapy.

Discussion

data show that the rate of bone de- is decreased about 40% in postmenopausal osteoporosis. Others have reported that patients with postmenopausal osteoporosis have calcium (17–19) and strontium (19, 20) kinetics indistinguishable from those of normal subjects. The small number of normal subjects reported in those studies limits comparison with our data.

A group comparable in size to ours was reported by Dymling (21). He measured ^{47}Ca kinetics in 24 normal subjects and in 34 patients with idiopathic osteopenia and found the patients had bone accretion rates 27% less than the normal subjects. Lower than normal values for strontium retention by osteoporotics have also been reported (22–24).

As has been stated (10), the decreased rate of bone deposition is not necessarily the cause of decreased skeletal mass but could be dependent on it. The rate of bone deposition per unit mass of bone may be normal or even above normal in the osteoporotic patients.

The nitrogen, phosphorus and calcium retention and general clinical improvement induced by androgen and/or estrogen treatment of patients with postmenopausal and senile osteoporosis are generally attri-

buted to stimulation of new bone matrix formation and its calcification (1). Stimulation of formation of new osteons that are in the process of mineralizing should be associated with increase in the rapidly miscible pool and in the rate of mineral deposition in bone. Such changes were not found in this study; in fact, the rate of bone deposition decreased in patients receiving estrogen therapy. Both androgen and estrogen therapy induced a decrease in urinary excretion rate. These findings suggest that in adults the calcium retention induced by androgens and estrogens results from a reduced rate of bone resorption instead of an increased rate of bone formation.

In the rapidly growing rat, large doses of estrogens prevent resorption of endosteal bone (25), and the reduced rate of resorption can be detected by kinetic analysis (26). Estrogen therapy also decreases the degree of bone resorption induced by injecting parathyroid extract into adult rats (27). Our data suggest a similar inhibitory effect of estrogens on bone resorption in the adult human skeleton. Using ^{45}Ca as a tracer, Haansy (28) has also found that treatment of three patients who had acute osteoporosis of disuse with "anabolic" steroids had no effect on bone deposition rate. Lafferty *et al.* (29) also concluded that bone resorption was probably decreased in four patients treated with androgens and estrogens.

In contrast, Dymling, Isaksson and Sjögren (30) found that in 14 patients 19-nortestosterone and 19-nortestosteronedecanoate had no effect on miscible pool size, bone deposition rate or urinary excretion rate as measured with ^{47}Ca. The difference between their results and the present data could stem from differences in the medication used, type of patient or method of data analysis. Their patients had osteopenia following gastric resection or in association with rheumatic disease. None of the patients in the present study had either of these conditions.

Although the bone resorption rate may be reduced after estrogen and androgen therapy, it might still be greater than, equal to, or less than the rate of bone deposition. In the first case, osteoporosis would progress more slowly than before therapy, in the second, it would be arrested, and in the last, bone mass would gradually increase. Many clinical observations suggest that osteoporosis can only be arrested by gonadal hormone therapy, and not reversed. One could therefore infer that resorption and deposition rates are equal after therapy and that these agents, previously thought to be anabolic for bone, are actually anticatabolic. The decreased bone deposition rate produced by estrogens may represent a secondary anti-anabolic effect.

It has been pointed out that isotopic bone deposition rate is the sum of new bone formation rates and bone crystal exchange rates (10). Therefore, the decrease in bone deposition caused by estrogen therapy could be due to decreased physicochemical reactivity of bone or to inhibition of cellular metabolism. These changes may not be specific since long-term high calcium intake has been reported to cause decrease in bone deposition rate (31, 32). Shorter periods of high calcium intake, like androgens, improved calcium balance without affecting bone deposition rate.

The present studies suggest that bone deposition rate is not primarily inhibited by corticoids. Hence, the decrease in bone mass that eventually occurs probably is the result of accelerated bone resorption. Increased bone resorption has been reported in other species when large doses of corticoids were given (33–35). Milhaud and associates (36) reported a decrease in *both* deposition and resorption rates in three three-month-old rats treated with corticoids for 21 days. No simultaneous control studies were reported, but the observations are probably valid and not in conflict with this report since corticoids interfere markedly with epiphysial cartilage development in growing animals.

The changes in renal excretion rate of strontium seen during steroid therapy could be secondary to an effect of altered bone metabolism on renal excretion of bone mineral or they could be caused by a direct renal effect of the steroids. Results of some studies in man have been interpreted as showing a direct effect of these agents on renal handling of calcium (37, 38), but the experimental designs of those studies and the present ones do not allow one to distinguish whether the primary site of action of steroids on calcium is at the skeleton or at the kidney or both.

Acknowledgments

The author is deeply indebted to Dr. Gilbert S. Gordan, who made available laboratory space in which the chemical determinations were made and who permitted study on some of his patients. Dr. Elmer Alpert and Mr. Glenn Watson helped supervise some of the clinical studies, and Mr. Warren Lubich provided expert technical assistance. Strontium gluconate was supplied by Dr. A. Carlotti and Mr. H. Althouse of Sandoz, Inc.

References

1. Albright, F., and E. C. Reifenstein, Jr., The Parathyroid Glands and Metabolic Bone Diseases: Selected Studies, Williams and Wilkins Co., Baltimore, 1948.
2. Honneman, P. H., and S. Wallach, *AMA Arch Intern Med* 100: 715, 1957.
3. Gardner, W. U., and C. A. Pfeiffer, *Physiol Rev* 23: 139, 1943.
4. Budy, A. M., M. R. Urist, and F. C. McLean, *Amer J Path* 28: 1143, 1952.
5. Wislocki, G. B., J. C. Aub, and C. M. Waldo, *Endocrinology* 40: 202, 1947.
6. Segaloff, A., and W. M. Cahill, *Proc Soc Exp Biol Med* 54: 182, 1943.
7. Kowalewski, K., *Endocrinology* 68: 759, 1958.
8. Baker, B. L., and D. J. Ingle, *Endocrinology* 48: 422, 1948.
9. Follis, R. H., Jr., *Bull Hopkins Hosp* 88: 466, 1951.
10. Eisenberg, E., and G. S. Gordan, *J Clin Invest* 40: 1809, 1961.
11. Eisenberg, E., *In* Pincus, G., and E. P. Vollmer (eds.), Biological Activities of Steroids in Relation to Cancer, Academic Press, New York, 1960, p. 189.
12. Eisenberg, E., E. Alpert, and G. S. Gordan, Hormonal Steroids I, Academic Press, Ltd., London, 1963, p. 252.
13. Likins, R. C., A. S. Posner, M. L. Knudo, and D. L. Craven, *Arch Biochem* 83: 472, 1959.
14. Comar, C. L., R. H. Wasserman, S. Ullberg, and G. A. Andrews, *Proc Soc Exp Biol Med* 95: 388, 1957.
15. Bauer, G. C. H., A. Carlsson, and B. Lindquist, *Acta Physiol Scand* 35: 56, 1955.
16. Aubert, J. P., F. Brunner, and L. J. Richelle, *J Clin Invest* 42: 885, 1963.
17. Heaney, R. P., and G. D. Whedon, *J Clin Endocr* 18: 1246, 1958.
18. Nordin, B. E. C., *Proc Roy Soc Med* 52: 361, 1959.
19. Dow, E. C., and J. B. Stanbury, *J Clin Invest* 39: 885, 1960.
20. Fraser, R., M. Harrison, and K. Ibbertson, *Quart J Med* 29: 85, 1960.
21. Dymling, J. F., *Acta Med Scand*, Suppl. 408, 1964.
22. Bauer, G. C. H., A. Carlsson, and B. Lindquist, *In* Fellinger, K., and H. Vetter (eds.), Radioaktive Isotope in Klinik und Forschung, Band III, Urban and Schwarzenberg, Munich, 1958, p. 25.
23. Spencer, H., M. Brothers, E. Berger, H. E. Hart, and D. Laszlo, *Proc Soc Exp Biol Med* 91: 156, 1956.
24. MacDonald, N. S., *Clin Orthop* 17: 154, 1960.
25. Urist, M. R., A. M. Budy, and F. C. McLean, *J Bone Joint Surg (Amer)* 32-A: 149, 1950.
26. Lindquist, B., A. M. Budy, F. C. McLean, and J. L. Howard, *Endocrinology* 66: 100, 1960.
27. Manuntа, G., J. Saroff, and C. W. Turner, *Proc Soc Exp Biol Med* 94: 785, 1957.
28. Heaney, R. P., *Amer J Med* 33: 186, 1962.
29. Lafferty, F. W., G. E. Spencer, and O. H. Pearson, *Amer J Med* 36: 514, 1964.
30. Dymling, J. F., B. Isaksson, and B. Sjögren, *In* Gross, F. (ed.), Anabolic Steroids in the Treatment of Osteopania in Protein Metabolism: Influence of Growth Hormone, Anabolic Steroids, and Nutrition in Health and Disease, Springer, Berlin, 1962, p. 412.
31. Schwartz, E., V. A. Panariello, and J. Saali, *J Clin Invest* 44: 1547, 1965.
32. Lutwak, L., *In* Pearson, O. H., and G. F. Joplin (eds.), Dynamic Studies of Metabolic Bone Disease, Blackwell Scientific Publications, Oxford, 1964, p. 87.
33. Clark, I., R. P. Geoffroy, and W. Bowers, *Endocrinology* 64: 848, 1959.
34. Cornett, E. R., R. L. Johnston, and E. J. Collins, *Pharm Sci* 51: 1050, 1962.
35. Urist, M. R., and N. M. Deutsch, *Endocrinology* 66: 805, 1960.
36. Milhaud, G., W. Ramagan, A. Gomes de Matos, and J. P. Aubert, *Rev Franc Etud Clin Biol* 5: 354, 1960.
37. Laaka, H., *Acta Endocr (Kobenhavn)* 34: 60, 1960.
38. Gardner, B., W. P. Graham, III, G. S. Gordan, H. F. Lokan, A. N. Thomas, and J. S. Teal, *J Clin Endocr* 23: 1115, 1963.

PATENT APPLICATION SERIAL NO. 07/963469

U.S. DEPARTMENT OF COMMERCE
PATENT AND TRADEMARK OFFICE
FEE RECORD SHEET

120 TD 11/03/92 07963469 1 201 355.00 CK

PTO-1556
(5/87)

A METHOD FOR AMELIORATING MUSCLE WEAKNESS/WASTING IN A PATIENT INFECTED WITH HUMAN IMMUNODEFICIENCY VIRUS-TYPE 1

Technical Field

The invention relates to the use of oxandrolone to attenuate myopathy and muscle weakness/wasting associated with infection by human immunodeficiency virus-Type 1.

Background of the Invention

Human immunodeficiency virus (HIV) associated myopathy and/or muscle weakness/wasting is a relatively common clinical manifestation of acquired immunodeficiency syndrome (AIDS). This is one of a number of neuromuscular disorders associated with the disease. There is some evidence to indicate that direct HIV infection of muscle may be at least partly responsible, occasionally resulting in a polymyositis-like disorder. In addition, zidovudine (AZT), an antiviral agent that is used widely in the clinical management of AIDS, has been associated with a toxic myopathy, presumably related to an inhibition of mitochondrial metabolism. In any event, the loss of muscle mass commonly observed in AIDS victims negatively impacts muscle function, however caused.

Individuals with HIV-associated myopathy or muscle weakness or wasting typically experience significant weight loss, generalized or proximal muscle weakness, tenderness, and muscle atrophy. Laboratory tests of samples from such individuals often reveal elevated levels of enzymes associated with muscle degeneration and necrosis, such as creatine kinase, aldolase, and aspartate amino transferase. Electromyographic test results for individuals with HIV-associated myopathy are typically consistent with

myopathic changes. Histopathologic tests may reveal muscle fiber necrosis associated with lymphocytic inflammatory infiltrates. In AZT myotoxicity, ragged red fibers are often observed.

Clinical management of HIV-associated myopathy and muscle weakness/muscle wasting varies. In individuals with AZT myopathy, withdrawal of this anti-retroviral agent may be associated with temporary improvement in strength and muscle bulk. Corticosteroid therapy, such as the administration of prednisone, has been occasionally successful when inflammatory infiltrates have been detected in muscle. However, a potential drawback to this approach is that corticosteroids, because of their immunosuppressant activity, may be harmful to individuals with AIDS who are already dangerously immunosuppressed as a consequence of the HIV infection.

Furthermore, corticosteroid use itself is associated with myopathies and an increased susceptibility to infections. Plasmapheresis has also been used with some success, although at least one patient has experienced, despite an increase in muscle strength, substantial weakness over a period of several weeks.

Summary of the Invention

The present invention provides a method which employs oxandrolone (an anabolic steroid with weak androgenic activity) as an alternative approach to the clinical management of HIV-associated myopathy/muscle weakness/muscle wasting. Loss in muscle mass (wasting) is attenuated, and body weight can be more readily maintained in this manner. Such an approach has been applied successfully to improve strength, reverse weight loss, and provide an improved sense of well-being.

Importantly, no evidence of liver injury or other untoward side effects have been observed.

Oxandrolone preferably is administered orally; however, other routes of administration can be utilized as well.

The present method of ameliorating muscle weakness or muscle wasting in a patient infected with HIV-1 comprises administering to the patient daily a sufficient amount of oxandrolone to attenuate the patient's rate of muscle mass loss. To this end, oxandrolone may be administered, orally or otherwise, in a daily dose in the range of about 2.5 to about 20 milligrams. However, the response of individual patients may vary and in some instances a daily dose greater than 20 mg may be required to achieve the desired response. The daily dose may be divided into unit doses of about 1 to about 5 milligrams each, administered to the patient three times per day at about eight-hour intervals.

<u>Detailed Description of the Preferred Embodiment</u>

Oxandrolone (17-hydroxy-17-methyl-2-oxaandrostan-3-one) is a known compound that is commercially available. The preparation of oxandrolone is described, inter alia, in U. S. Patent No. 3,128,283 to Pappo, which description is incorporated herein by reference.

Pharmacologically, oxandrolone is a synthetic anabolic steroid similar in structure to testosterone, but having a different, lesser androgenic/anabolic activity ratio. In addition, oxandrolone is unique among all other testosterone analogues in that it contains an oxygen atom instead of a methylene group at the 2-position of the phenanthrene nucleus. In addition, oxandrolone lacks a 4-ene function in its A-ring. The anabolic potency of oxandrolone, estimated as

approximately 3 to 13 times that of testosterone, is believed to result form this unique structure.

Oxandrolone disposition and metabolism in man has been studied following oral administration of a 10 milligram dose. The study indicated that oxandrolone was rapidly and completely absorbed, yielding a mean peak plasma concentration of 417 micrograms of oxandrolone per milliliter at 66 minutes. The plasma concentration of oxandrolone declined in a biphasic manner with a distribution half-life of approximately 30 minutes and an elimination half-life of 9.4 hours. Protein binding of oxandrolone was observed to be extensive.

In distinct contrast to other anabolic androgenic steroids such as methyltestosterone, fluoxymesterone, and micronized testosterone, oxandrolone taken orally is excreted mainly unchanged and unconjugated in urine. Urinary excretion of approximately 35 percent of an oral oxandrolone dose has been observed within 72 hours after ingestion. After 96 hours, approximately 65 percent of the administered oxandrolone dose was excreted in urine. Fecal excretion accounts for less than about 3 percent over the same time period.

Oxandrolone compositions, upon administration in accordance with this invention, ameliorate myopathy and muscle weakness in patients suffering from infections by human immunodeficiency virus-Type 1. Anabolic steroids, as a class, are known to stimulate appetite. Improved nutrition is important to individuals with AIDS who have experienced loss of lean body mass. Further, as a consequence of direct interaction with androgen and/or glucocorticoid receptors in muscle, anabolic steroids promote muscle

anabolism through both anabolic pathways and anticatabolic pathways.

Anabolic steroids, such as oxandrolone, also increase protein synthesis. For example, oxandrolone increased muscle protein synthesis in a study of acute uremic rats. Similarly, administration of oxandrolone preceded clinical improvement in appetite, cell mass, linear growth, and weight for height in boys with chronic renal failure. These observations are consistent with anabolic activity. Oxandrolone may also stimulate the secretion of growth hormone and insulin-like growth factors.

In addition to producing beneficial direct anabolic action, oxandrolone is also believed to act as a delayed immunostimulant. In contrast, other appetite stimulants, such as dronabinol, that are currently under evaluation as appetite stimulants for AIDS patients can act as immunosuppressants in animals.

For purposes of administration in accordance with this invention, the active ingredient oxandrolone is combined with solid or liquid pharmaceutical carriers and formulated in unit dosage form using pharmacologically acceptable excipients, or dissolved or suspended in physiologically acceptable solvents or liquid vehicles for oral, percutaneous, or topical administration.

The overall daily dose of oxandrolone to provide a therapeutically effective amount in accordance with the method of this invention can be as low as about 2.5 milligrams and as high as about 20 milligrams, depending upon the patient's response and the mode of administration.

The amount of the active ingredient within the aforementioned ranges that is to be administered depends upon the age, weight and condition of the patient, as

well as on factors such as the frequency and route of administration. In formulating oxandrolone, it is recognized that there may be differences between the immediate and the long term response. To account for these changes, the specific dosage given to a particular patient is based also on the individual patient's response. Preferably, oxandrolone is orally administered to the patient daily for a time period in the range of about 2 weeks to about 6 months.

Attenuation of the rate of muscle mass loss in a patient can be ascertained by comparing the patient's rate of weight loss before oxandrolone therapy with that after the administration of oxandrolone has been commenced. Alternatively, or in addition, the patient's urinary nitrogen level can be monitored, a well-known expedient. A decrease in the patient's urinary nitrogen level is indicative of a decrease in muscle mass loss.

Similarly, the maintenance of a relatively stable patient's total body potassium level, as well as an increase in the patient's total body potassium level, upon oxandrolone administration indicates that a therapeutically effective amount of oxandrolone is being administered. A patient's total body potassium level can be monitored, for example, as described in Kotler et al., **The American Journal of Clinical Nutrition**, 42:1255-1265 (December 1985) and Pierson, Jr., et al., **Am. J. Physiol.**, 246 (Renal Fluid Electrolyte Physiol. 15):F234-F239 (1984).

The route of administration can be oral, percutaneous, transdermal, sublingual, buccal, intravenous, intramuscular, or the like. Of these, oral administration is preferred. The patient's daily dose of the active ingredient preferably is in the range of about 7.5 milligrams, but may exceed 20 milligrams based on clinical response. This daily dose can be given in

tablet form as a single dose, or as plural divided doses, preferably 2 to 3 divided doses. The requisite daily dose can also be supplied continuously, for example, by a transdermal patch worn by the patient or intravenously. If the oxandrolone is administered orally, dosages in the range of about 2 to about 5 milligrams three to four times daily typically may be utilized.

Oxandrolone tablets are manufactured using standard solid dose form technology in accordance with United States Pharmacopeia (USP) specifications (see, for example, The United States Pharmacopeia, 22nd Revision, pp. 981-982). Specifically, a typical 150-milligram tablet contains the following:

Oxandrolone, USP	2.5 mg
Corn Starch, NF	30.0 mg
Lactose NF (hydrous)	113.0 mg
Hydroxypropyl Methylcellulose, USP	3.0 mg
Magnesium Stearate	1.5 mg
	150.0 mg

The terms "unit dosage form" and "unit dose" as used in the present specification and claims refer to a physically discrete unit or units suitable as unitary doses for patients, each unit containing a predetermined quantity of the active ingredient calculated to produce the desired therapeutic effect in association with the pharmacologically acceptable carrier. The specifications for the unit dosage forms of this invention are dictated in part and are also dependent upon (a) the unique characteristics of the active ingredient and (b) the particular therapeutic effect to be achieved, as well as upon limitations inherent in the art of compounding such active ingredient for the therapeutic use disclosed in detail in this specification. Examples of suitable unit dosage forms

in accordance with this invention are tablets, pills, powder packets, wafers, cachets, segregated multiples of any of the foregoing, transdermal patches, aliquots of injectables, and the like forms.

The primary response variables are patient's total body potassium, body weight, muscle mass, muscle strength, improvement in or increased appetite, and general sense of well-being. In addition, improvement in immune status (or at a minimum, no worsening of immune function) in response to oxandrolone is significant as well.

An important question regarding the use of any drug in combination with anti-retroviral therapy is whether drug interactions may occur that would diminish AZT efficacy or increase the frequency of severity of AZT-related adverse reactions. TABLE 1 compares various published pharmacological parameters for oxandrolone and AZT and illustrates important differences between the two drugs.

TABLE 1

Comparison of Selected Oxandrolone and AZT Pharmacology Parameters

Parameter	Oxandrolone	AZT
Oral Bioavailability	100%	65%
Tmax	1.1 hr	0.7 hr
Biological T1/2	9.4 hr	1.1 hr
Vd	578 ml/kg	>1400 ml/kg
Protein Binding	>95%	25-35%
Plasma Clearance	43 ml/kg/hr	>1300 ml/kg/hr
Metabolism	Little	Extensive
Glucuronidation	Little	Substantial
Urinary Excretion	Extensive; primarily parent compound	Extensive; parent and glucuronide conjugated
Target Organ Toxicity	Liver (anabolic steroids as a class)	Hematopoietic system (e.g., anemia, granulocytopenia)
Known Drug Interactions	Anticoagulants; oral hypoglycemic agents; adrenal steroid when edema present	Drugs that may: (a) inhibit glucuronidation (e.g., aspirin, acetaminophen) or urinary excretion (e.g., probenecid); (b) adversely affect blood cell number and function; and (c) nephrotoxic or cytotoxic

Because oxandrolone is primarily protein bound, whereas AZT is primarily non-protein bound, oxandrolone will not compete appreciably with AZT for binding sites in plasma. Consequently, administration of oxandrolone to patients on AZT therapy is unlikely to alter the level of free AZT in the blood. Likewise, the administration of AZT is unlikely to alter the level of free oxandrolone in the blood. An oxandrolone-AZT drug interaction involving binding site displacement is, therefore, extremely unlikely.

AZT is rapidly metabolized and excreted in the urine--a significant quantity is excreted in the form of glucuronide conjugates. In sharp contrast, oxandrolone, perhaps due to presence of a lactone group and the absence of a 4-ene function in the A-ring, undergoes little hepatic metabolism and is excreted primarily unchanged and unconjugated in urine. Thus, in contradistinction to other drugs that may competitively inhibit glucuronidation and thereby potentially slow the rate of AZT metabolism, such as aspirin, acetaminophen, or indomethacin, the present active agent, oxandrolone, is not believed to affect AZT metabolism.

Furthermore, oxandrolone is neither nephrotoxic nor cytotoxic. Accordingly, oxandrolone is not expected to interfere with the renal excretion of AZT or its metabolites. To the contrary, oxandrolone has been safely and effectively used in patients with chronic renal disease to stimulate growth and increase lean body mass. In well-controlled studies of oxandrolone for the clinical management of critically ill patients with acute alcoholic hepatitis, oxandrolone administered at daily doses of up to 80 mg/day for four weeks and 40 mg/day for eight weeks did not result in any drug-related nephrotoxicity.

While it is known that anabolic androgenic steroids have been associated with potentially life-threatening forms of liver disease, including peliosis hepatis, cholestatic jaundice, and hepatocellular neoplasms, specific reports in the medical literature regarding liver disease in oxandrolone-treated patients, at the dosages proposed for use in the clinical management of HIV associated muscle weakness/wasting (i.e., about 2.5 to about 20 mg/day) are rare.

Oxandrolone and AZT have different mechanisms of action. They also function in different sites of cellular action at the receptor level. Oxandrolone functions via interaction with androgen and glucocorticoid receptors, whereas AZT, once phosphorylated, acts to inhibit HIV reverse transcription. Thus, competitive inhibition of AZT by oxandrolone at the cellular level also is considered unlikely.

Neither has oxandrolone been associated with anemia or granulocytopenia, two frequently occurring and potentially serious side effects associated with AZT therapy. To the contrary, anabolic androgenic steroids have been used clinically to stimulate ethyropoiesis in hypoanemias, aplastic anemias, hemolytic anemias, renal anemias, anemias due to cytotoxic therapy, and various leukemias. It has been reported recently that androgens augment beneficial effects of erythropoietin in the treatment of anemia resulting from end-stage renal disease.

Data derived from animal models and human clinical studies indicate that anabolic steroids are unlikely to suppress immune function in patients infected with HIV. For example, anabolic steroids can stimulate granulopoiesis in mice, as evidenced by stimulation of granulocytic colony-forming cells derived

macrophage activity and cell-mediated immunity in patients with uterine cervical cancer when administered parentally. In related studies, anabolic steroids increased peripheral lymphocyte and monocyte counts, Immunoglobin G (IgG) levels, and PHA-blastoid transformation of peripheral lymphocytes. In those studies, β_2-microglobulin levels simultaneously decreased.

IgG is one of a class of antibodies secreted by B cells (i.e., B-lymphocytes) in response to an antigenic challenge (e.g., foreign protein like that from bacteria). In the case of HIV infection, humoral immune function (i.e., B-cell mediated) is significantly impaired. Accordingly, when HIV-infected individuals are challenged with a specific antigen, the typical response of B-cell proliferation, differentiation and secretion of antibodies (e.g., IgG) is diminished or absent. This decline in humoral immune function coupled with defects in cellular immune (i.e., T-cell) function contributes to the overall failure of the immune system to respond in an appropriate manner to challenge. B-cells in AIDS victims are, by mechanisms unknown, hyperstimulated to secrete large amounts of immunoglobulins that make the humoral system refractory to new antigens. The result is that the patient's system no longer recognizes new antigens and does not respond.

In animal studies in which anabolic steroids have been reported to increase IgG and PHA-blastoid activity, these changes occurred as a result of immune system stimulation, and are positive responses. β-microglobin is a cell surface protein that is found on all nucleated cells and it is released into the serum

during cell turnover. Generally, β-microglobulin is considered a marker of infectious, inflammatory, malignant and autoimmune disease activity. In several AIDS studies, β-microglobulin levels correlated with disease progression and T4 (T-helper) cell counts. In the case of therapy with oxandrolone, for example, a decrease in β-microglobulin levels is desirable. Thus, animal data showing reduced plasma levels of β-microglobulin in response to anabolic steroids is evidence of a positive effect and suggestive of similar activity in man.

Accordingly, there are no reasons to believe that the administration of an anabolic steroid in general and oxandrolone in particular would have adverse effects on the immune system. Generally, the target organ of toxicity for these drugs is the liver--probably because this is where most are metabolized. Oxandrolone, however, has a remarkably good safety profile in man as a likely consequence of its resistance to hepatic metabolism; an oral dose is excreted primarily in urine as the parent compound, as stated hereinabove.

Data from clinical trials in patients with severe alcoholic liver disease provide further evidence that oxandrolone is not likely to suppress immune function in patients with HIV infection. Ethanol abuse is associated with loss of lymphocyte functions, particularly T-cell dependent immune responses. Previous researchers have observed that oxandrolone significantly improved lymphocyte number in patients with severe alcoholic hepatitis. Because the loss of lymphocytic function by alcoholic liver disease parallels, to a significant degree, the loss of T-cell function due to HIV infection, it is reasonable to

hypothesize that oxandrolone will increase the T-Cell function of HIV-infected patients.

Therefore, these data from laboratory animals and human studies indicate that suppression of the immune system by anabolic steroids, such as oxandrolone, is unlikely. Nonetheless, subjects undergoing oxandrolone therapy, as a precaution, should be monitored for changes in lymphocyte number, particularly CD4+ and CD8+, as is routinely done for patients who undergo steroid therapy.

In summary, based on the differences between AZT and oxandrolone with respect to pharmacokinetics, metabolism, reported drug interactions, mechanisms of action, and reported toxicities, oxandrolone and AZT can be safely used in combination for subjects infected with the Type-1 HIV virus and suffering from HIV-associated myopathy. The use of oxandrolone in patients on AZT therapy is, on the basis of known drug interactions, also consistent with current FDA-approved labeling for AZT and oxandrolone.

The following example demonstrates the effectiveness of oxandrolone in attenuating the effects of HIV-associated muscle weakness or muscle wasting in an AIDS patient.

EXAMPLE

A patient, a thirty-two year old homosexual man, known to be HIV-seropositive since February 1989, noted difficulty opening drawers and bottles in May 1989. The patient weakened progressively and, during a physical examination in September 1989, demonstrated by confrontation testing the weakness of neck flexion and proximal limbs. However, his muscle stretch reflexes remained normal. Laboratory tests showed the patient's creatine kinase level to be 286 International Units per

liter, much higher than the normally observed range for creatine kinase of about 40-200 Units per liter.

Zidovudine (azidothymidine or AZT) was initiated at 500 milligrams daily, but the patient's strength continued to decline through February 1990. He complained of an inability to ascend a flight of stairs. The patient exhibited greater weakness and atrophy of neck flexors and extremity muscles during another physical examination performed at this time. An electromyogram revealed a decrease of amplitude and duration of the patient's motor unit potentials and increased recruitment in selected muscles of his right upper extremity. The patient's creatine kinase tested at 456 Units per liter. A muscle biopsy revealed numerous myofibers, abundant ragged red fibers, and numerous eosinophilic inclusions. Round cell inflammatory infiltrates were also noted. In light of these developments, the zidovudine treatment was terminated.

Substantial improvement initially followed the discontinuation of zidovudine. However, because of a subsequent continued and progressive weakness rendering it difficult for the patient to ascend or descend a flight of stairs, a prednisone therapy (60 mg daily) was initiated. No significant improvement accompanied the use of prednisone.

Thereafter, a trial period of oral oxandrolone administration (2.5 milligrams, three times daily, in tablet form) was initiated. Within two weeks of the initiation of the oxandrolone therapy, the patient noted an improved sense of well being, became stronger, and gained weight. Within one month, he was able to ascend and descend stairs without problems. Confrontation testing revealed nearly normal strength. The patient's weight increased from 115 pounds to 130 pounds. The

patient's muscle atrophy was alleviated as well. Liver functions were closely monitored for signs of elevation, but undesirable side effects were not detected.

After several months of the aforementioned therapy with oxandrolone, the patient was no longer able to obtain oxandrolone for use as a medication. Weakness and weight loss ensued. Trials of other anabolic preparations, specifically stanazol and oxymethalone, did not return the patient to his previous levels of function and strength.

The EXAMPLE demonstrates that oxandrolone can be a beneficial alternative for clinical management of HIV-associated myopathy and muscle weakness and wasting.

It is intended that the foregoing description is by way of illustration only and is not to be construed as limiting the invention in any way except in the spirit and scope of the appended claims.

WHAT IS CLAIMED IS:

1. A method for ameliorating myopathy and muscle weakness in a patient infected with a Type-1 human immunodeficiency virus which comprises administering to said patient oxandrolone in an amount sufficient to attenuate the rate of muscle mass loss in said patient.

2. The method in accordance with claim 1 wherein the oxandrolone is administered to said patient in a daily dosage in the range of about 2.5 to about 20 milligrams.

3. The method in accordance with claim 2 wherein the daily dosage of the oxandrolone is about 7.5 milligrams.

4. The method in accordance with claim 1 wherein the oxandrolone is administered to said patient as a unit dose of about 1 to about 5 milligrams three times per day at about eight-hour intervals.

5. The method in accordance with claim 1 wherein the oxandrolone is administered percutaneously.

6. The method in accordance with claim 1 wherein the oxandrolone is administered intravenously.

7. The method in accordance with claim 1 wherein the oxandrolone is administered intramuscularly.

8. The method in accordance with claim 1 wherein the oxandrolone is administered sublingually.

9. The method in accordance with claim 1 wherein the oxandrolone is administered transdermally.

10. The method in accordance with claim 1 wherein the oxandrolone is administered orally.

11. The method in accordance with claim 10 wherein the oxandrolone is administered in the form of a tablet.

12. The method in accordance with claim 1 wherein the administration is continued over a period in the range of about 2 weeks to about 6 months.

13. A method for ameliorating HIV-associated myopathy and muscle wasting in a patient infected with a Type-1 human immunodeficiency virus which comprises orally administering a therapeutically effective amount of oxandrolone to said patient daily for a time period in the range of about 2 weeks to about 6 months.

Abstract of the Disclosure

A method for attenuating the HIV-associated myopathy and muscle wasting associated with infection by human immunodeficiency virus-Type 1. Administration of oxandrolone in a daily dosage of about 2.5 to about 20 milligrams is described.

Dkt. 44657-AAA-PCT-US/JPW/GJG

IN THE UNITED STATES PATENT OFFICE

Applicant	:	Joseph R. Berger
U.S. Serial No.	:	10/052,961
Filed	:	January 18, 2002
For	:	A METHOD FOR AMELIORATING MUSCLE WEAKNESS/WASTING IN A PATIENT INFECTED WITH HUMAN IMMUNODEFICIENCY VIRUS-TYPE 1

1185 Avenue Of The Americas
New York, New York 10036
March 5, 2004

Mail Stop OIPE
Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

Sir:

REQUEST TO CORRECT ERROR IN FILING RECEIPT

This communication is filed to request the issuance of a corrected Filing Receipt in connection with the above-identified application. Upon receipt of the official Filing Receipt for the subject application, a copy of which is attached hereto as **Exhibit A**, applicants' undersigned attorney noticed an error.

Specifically, the Domestic Priority data is listed incorrectly.

A corrected filing receipt should read as follows:

--Domestic Priority data

THIS APPLICATION IS A CON OF 09/469,817 12/22/1999 **PAT 6,670,351**
WHICH IS A CON OF 08/244,988 06/22/1995 PAT 6,090,799
WHICH IS A 371 OF PCT/US93/10063 10/20/1993
WHICH IS A CON OF 07/963,469 10/20/1992--

Applicants contend that the correct data may be found in the Declaration and Power of Attorney which was filed in the grand-parent of the subject application, U.S. Serial 08/244,988. A

copy of the Declaration and Power of Attorney is provided herewith as **Exhibit B**. Accordingly, applicants request that a corrected Filing Receipt be issued.

No fee is deemed necessary in connection with the filing of this Request. However, if any fee is required, authorization is hereby given to charge the amount of any such fee to Deposit Account 03-3125.

Respectfully submitted,

Gary J. Gershik

John P. White
Registration No. 28,678
Gary J. Gershik
Registration No. 39,992
Attorneys for Applicants
Cooper & Dunham LLP
1185 Avenue of the Americas
New York, New York 10036
(212) 278-0400

I hereby certify that this correspondence is being deposited this date with the U.S. Postal Service with sufficient postage as first class mail in an envelope addressed to: Mail Stop OIPE, Commissioner for Patents, P.O. Box 1450, Alexandria, VA 22313-1450

Gary J. Gershik 3/5/04

John P. White Date
Reg. No. 28,678
Gary J. Gershik
Reg. No. 39,992

JPW

UNITED STATES PATENT AND TRADEMARK OFFICE

COMMISSIONER FOR PATENTS
UNITED STATES PATENT AND TRADEMARK OFFICE
WASHINGTON, D.C. 20231
www.uspto.gov

APPLICATION NUMBER	FILING DATE	GRP ART UNIT	FIL FEE REC'D	ATTY.DOCKET.NO	DRAWINGS	TOT CLAIMS	IND CLAIMS
10/052,961 ✓	01/18/2002 ✓	2855	370	44657-AAA-PCT-US/JPW		16	2

CONFIRMATION NO. 3958

FILING RECEIPT

OC000000007456216

John P. White
Cooper & Dunham LLP
1185 Avenue of the Americas
New York, NY 10036

RECEIVED
FEB 14 2002
W.D.

Date Mailed: 02/11/2002

Receipt is acknowledged of this nonprovisional Patent Application. It will be considered in its order and you will be notified as to the results of the examination. Be sure to provide the U.S. APPLICATION NUMBER, FILING DATE, NAME OF APPLICANT, and TITLE OF INVENTION when inquiring about this application. Fees transmitted by check or draft are subject to collection. Please verify the accuracy of the data presented on this receipt. **If an error is noted on this Filing Receipt, please write to the Office of Initial Patent Examination's Customer Service Center. Please provide a copy of this Filing Receipt with the changes noted thereon. If you received a "Notice to File Missing Parts" for this application, please submit any corrections to this Filing Receipt with your reply to the Notice. When the USPTO processes the reply to the Notice, the USPTO will generate another Filing Receipt incorporating the requested corrections (if appropriate).**

Applicant(s)

Joseph R. Berger, Miami, FL;

Assignment For Published Patent Application

BTG Pharmaceutical Corp.;

Domestic Priority data as claimed by applicant

THIS APPLICATION IS A CON OF 09/469,817 12/22/1999
WHICH IS A CON OF 08/244,988 06/22/1995 PAT 6,090,799
WHICH IS A 371 OF PCT/US93/10063 10/20/1993

Foreign Applications

If Required, Foreign Filing License Granted 02/11/2002

Projected Publication Date: 05/23/2002

Non-Publication Request: No

Early Publication Request: No

**** SMALL ENTITY ****

Title

Method for ameliorating muscle weakness/wasting in a patient infected with human immunodeficiency virus-type 1

Preliminary Class

073

LICENSE FOR FOREIGN FILING UNDER
Title 35, United States Code, Section 184
Title 37, Code of Federal Regulations, 5.11 & 5.15

GRANTED

The applicant has been granted a license under 35 U.S.C. 184, if the phrase "IF REQUIRED, FOREIGN FILING LICENSE GRANTED" followed by a date appears on this form. Such licenses are issued in all applications where the conditions for issuance of a license have been met, regardless of whether or not a license may be required as set forth in 37 CFR 5.15. The scope and limitations of this license are set forth in 37 CFR 5.15(a) unless an earlier license has been issued under 37 CFR 5.15(b). The license is subject to revocation upon written notification. The date indicated is the effective date of the license, unless an earlier license of similar scope has been granted under 37 CFR 5.13 or 5.14.

This license is to be retained by the licensee and may be used at any time on or after the effective date thereof unless it is revoked. This license is automatically transferred to any related applications(s) filed under 37 CFR 1.53(d). This license is not retroactive.

The grant of a license does not in any way lessen the responsibility of a licensee for the security of the subject matter as imposed by any Government contract or the provisions of existing laws relating to espionage and the national security or the export of technical data. Licensees should apprise themselves of current regulations especially with respect to certain countries, of other agencies, particularly the Office of Defense Trade Controls, Department of State (with respect to Arms, Munitions and Implements of War (22 CFR 121-128)); the Office of Export Administration, Department of Commerce (15 CFR 370.10 (j)); the Office of Foreign Assets Control, Department of Treasury (31 CFR Parts 500+) and the Department of Energy.

NOT GRANTED

No license under 35 U.S.C. 184 has been granted at this time, if the phrase "IF REQUIRED, FOREIGN FILING LICENSE GRANTED" DOES NOT appear on this form. Applicant may still petition for a license under 37 CFR 5.12, if a license is desired before the expiration of 6 months from the filing date of the application. If 6 months has lapsed from the filing date of this application and the licensee has not received any indication of a secrecy order under 35 U.S.C. 181, the licensee may foreign file the application pursuant to 37 CFR 5.15(b).

Declaration and Power of Attorney

As a below-named inventor, I hereby declare that:

My residence, post office address and citizenship are as stated below next to my name.

I believe I am the original, first and sole inventor (if only one name is listed below) or an original, first and joint inventor (if plural names are listed below) of the subject matter which is claimed and for which a patent is sought on the invention entitled:

A METHOD FOR AMELIORATING MUSCLE WEAKNESS/WASTING IN A PATIENT INFECTED WITH HUMAN IMMUNODEFICIENCY VIRUS-TYPE 1

the specification of which
(check one)

____ is attached hereto.

_X_ was filed on October 20, 1993 as

Application Serial No. PCT/US93/10061; U.S. Serial No. 08/244,988

and was amended on _____
(if applicable)

I hereby state that I have reviewed and understand the contents of the above-identified specification including the claims, as amended by any amendment referred to above.

I acknowledge the duty to disclose information of which I am aware which is material to the examination of this application in accordance with Title 37, Code of Federal Regulations Section 1.56(a).

I hereby claim foreign priority benefits under Title 35, United States Code, Section 119 of any foreign application(s) for patent or inventor's certificate listed below and have also identified below any foreign application for patent or inventor's certificate having a filing date before that of the application on which priority is claimed:

Prior Foreign Application(s) Number	Country	Filing Date	Priority Claimed Yes	No
NONE				

I hereby claim the benefit under Title 35, United States Code Section 120 of any United States Application(s) listed below and, insofar as the subject matter of each of the claims of this application is not disclosed in the prior United States application in the manner provided by the first paragraph of Title 35, United States Code, Section 112, I acknowledge the duty to disclose material information as defined in Title 37, Code of Federal Regulations Sections 1.56(a) which occurred between the filing date of the prior application and the national or PCT international filing date of this application:

Application Serial No.	Filing Date	Status
PCT/US93/10063	October 20, 1993	
07/963,469	October 20, 1992	

And I hereby appoint

John P. White (Reg. No. 28,678); Norman H. Zivin (Reg. No. 25,385); Thomas F. Moran (Reg. No. 16,579); Ivan S. Kavrukov (Reg. No. 25,161); Christopher C. Dunham (Reg. No. 30,141); Peter J. Phillips (Reg. No. 29,691); Richard S. Milner (Reg. No. 33,970); Matthew J. Golden (Reg. No. 35,161); Albert Wai-Kit Chan (Reg. No. 36,479); Matthew B. Tropper (Reg. No. 37,457); Lewis J. Kreisler (Reg. No. 38,522); Robert T. Maldonado (Reg. No. 38,232).

and each of them, all c/o Cooper & Dunham LLP, 1185 Avenue of the Americas, New York, New York 10036, my attorneys, each with full power of substitution and revocation, to prosecute this application, to make alterations and amendments therein, to receive the patent, to transact all business in the Patent and Trademark Office connected therewith and to file any International Applications which are based thereon under the provisions of the Patent Cooperation Treaty.

Please address all communications, and direct all telephone calls, regarding this application to

John P. White Reg. No. 28,678

Cooper & Dunham LLP
1185 Avenue of the Americas
New York, New York 10036
Tel. (212) 278-0400

I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

Full name of sole or first joint inventor Joseph R. Berger (Sim Fass, on behalf of and as agent for)

Inventor's signature

Citizenship United States of America Date of signature June 21, 1995

Residence 6460 S.W. 109th Street, Miami, Florida 33156

Post Office Address University of Kentucky-Annex 4, Chambers Building, Room 228E
Lexington, Kentucky 40536

Signed by Sim Fass, an authorized official of the assignee, BTG Pharmaceutica Corp., on behalf of and as agent for Joseph R. Berger.